

What is claimed is:

1. (Original) A method for extracting intact cytoplasmic biomolecules from cells, which method comprises:
 - a. obtaining a fluid sample from a test subject, said fluid sample comprising a mixture of cell populations suspected of containing target cells;
 - b. releasing said cytoplasmic biomolecules from said cells;
 - c. isolating said cytoplasmic biomolecules; and
 - d. analyzing said cytoplasmic biomolecules.
2. (Original) The method as claimed in claim 1, wherein said fluid sample is whole blood.
3. (Currently Amended) The method as claimed in claim 1, wherein selection of said target cells is accomplished by ~~a selecting method from the group consisting of immunomagnetic selection, cell fractionation, and combinations thereof.~~
4. (Original) The method according to claim 1, wherein said releasing step is accomplished by addition of a permeabilizing agent.
5. (Original) The method according to claim 4, wherein said permeabilizing agent is selected from the group consisting of a detergent, surfactant, and combinations thereof.
6. (Original) The method according to claim 4, wherein said permeabilizing agent is selected from the group consisting of saponin, Immuniperm, and combinations thereof.
7. (Cancel) The method according to claim 4, wherein said permeabilizing agent is Immuniperm.
8. (Original) The method according to claim 1, wherein said obtaining said fluid sample includes treating the sample with a cell stabilizing agent.
9. (Currently Amended) The method according to claim 8, wherein said stabilizing agent is ~~selected from the group consisting of aldehydes, urea, and combinations thereof.~~
10. (Cancel) The method according to claim 9, wherein said aldehyde is selected from the group consisting of formaldehyde, paraformaldehyde, and combinations thereof.

11. (Cancel) The method according to claim 8, wherein said stabilizing agent is a dialdehyde.
12. (Cancel) The method according to claim 11, wherein said dialdehyde is selected from the group consisting of glutaraldehyde, glyoxal, and combinations thereof.
13. (Original) The method according to claim 8, wherein said releasing of said cytoplasmic biomolecules involves macromolecular complexes formed after exposure to said stabilizing agent.
14. (Original) The method according to claim 13, wherein said releasing said cytoplasmic biomolecules from said cells is accomplished with enzymatic digestion.
15. (Currently Amended) The method according to claim 14, wherein said enzymatic digestion is accomplished with proteinase K digestion ~~a selection from the group of proteinases, nucleophiles, and combinations thereof.~~
16. (Cancel) The method according to claim 15, wherein said proteinases are selected from the group consisting of proteinase K digestion, V8 proteinase digestion, pronase digestion, and combinations thereof.
17. (Original) The method according to claim 15, wherein said nucleophiles are from a group consisting of phosphate-based buffers, tris-based buffers, acetic hydrazide, hydroxylamine, and combinations thereof.
18. (Cancel) The method according to claim 1, wherein said cytoplasmic biomolecules are proteins.
19. (Cancel) The method according to claim 18, wherein said isolating said proteins is accomplished by a method selected from the group consisting of chemical extractions, electrophoresis, chromatography, immunoseparations and affinity techniques.
20. (Original) The method according to claim 1, wherein said cytoplasmic biomolecules are nucleic acids.
21. (Currently Amended) The method according to claim 20, wherein said nucleic acids are selected from ~~the group consisting of cytoplasmic RNA, nuclear and mitochondrial RNA, nuclear and mitochondrial DNA, and combinations thereof.~~

22. (Cancel) The method according to claim 20, wherein said nucleic acids are cytoplasmic mRNA.
23. (Original) The method according to claim 20, wherein the method of isolating said nucleic acids is selected from the group consisting of RNA or DNA chemical extractions, electrophoresis, chromatography, immunoseparations and affinity techniques.
24. (Original) The method according to claim 20, wherein said isolating of said nucleic acids is accomplished by capture with magnetic beads affixed to oligo(dT).
25. (Original) The method according to claim 1, wherein said target cells are assessed for phenotypic expression other than with said cytoplasmic biomolecules after obtaining said fluid sample.
26. (Currently Amended) The method according to claim 25, wherein said phenotypic expression is selected by ~~from the group consisting of~~ morphological examining, ~~cell component staining, immunoanalyzing, and combinations thereof.~~
27. (Cancel) The method according to claim 1, wherein said analysis of said cytoplasmic biomolecules is accomplished by functional proteomics.
28. (Cancel) The method according to claim 27, wherein said functional proteomics is selected from the group consisting of protein expression profile, Western blot, amino acid sequence analysis, electrophoresis, 2-D electrophoresis, mass spectrometry, gas chromatography, liquid chromatography nuclear magnetic resonance, infrared, atomic adsorption, and combinations thereof.
29. (Cancel) The method according to claim 1, wherein said analyzing of said cytoplasmic biomolecules is accomplished by functional genomics.
30. (Cancel) The method according to claim 29, wherein said functional genomics is selected from the group consisting of mRNA profile analysis, protein expression profile analysis, polymerase chain reaction, Northern blot, Western blot, Nucleotide or amino acid sequence analysis, gene expression on a microarray, electrophoresis, 2-D electrophoresis, mass spectrometry, gas chromatography, liquid chromatography nuclear magnetic resonance, infrared, and atomic adsorption.

31. (Original) The method according to claim 1, wherein said analyzing of said cytoplasmic RNA for the presence of at least two genetic markers is accomplished by multigene RT-PCR.
32. (Original) The method according to claim 1, wherein said analyzing of said cytoplasmic RNA for said genetic markers comprises the further steps of:
- a. reverse transcribing said genetic markers with at least one set of gene specific primers all of which have a universal primer extension on the 5' terminus;
 - b. removing said gene specific primers;
 - c. amplifying a gene specific amplicon with said universal primer extensions in a PCR amplification; and
 - d. identifying said gene specific amplicon.
33. (Original) The method according to claim 32, wherein said removing of said gene specific primers is accomplished using a method selected from the group consisting of molecular size exclusion, solid support selective attachment, and single strand specific DNase, and thereafter incorporating uracil-N-glycosylase with DNA oligonucleotide primers synthesized with deoxyUridine.
34. (Original) The method according to claim 32, wherein said removing of said gene specific primers is accomplished with uracil-N-glycosylase treatment of DNA oligonucleotide primers synthesized with deoxyUridine.
35. (Original) The method according to claim 34, wherein said uracil-N-glycosylase treatment is followed by incorporation of DNase-free RNases.
36. (Original) The method according to claim 33, wherein said gene specific primers are used under conditions of high primer-target annealing specificity.
37. (Original) The method according to claim 36, wherein said high primer-target annealing specificity is accomplished with proteins from natural recombination cellular repair mechanisms.
38. (Original) The method according to claim 37, wherein said high primer-target annealing specificity is accomplished with recA.

Claims 39-186 (Cancel)